AP Biology Lab Summary of Key Points

Be Aware of the Following for Most Investigations

- What are the controls?
  - What do you need to keep constant? How?
- What variables can you test?
  - Test ONE at a time
  - How will you test them?
  - What is the independent vs dependent variable?
- What are possible sources of error and how can they potentially be corrected?
- Be able to create and interpret graphs
  - Title
  - Label axes
  - Choose EQUAL intervals when numbering axes
  - Calculate rates ($\Delta y/\Delta x$), mean (average), etc.

Investigation 1: Artificial Selection

- Need **variation** for natural or artificial selection to occur
  - Ex: Trichome density
    - **Trichomes** are tiny hair like structures on plant shoots
    - Defend against herbivores and pathogens by forming a mechanical barrier or secreting chemicals
- Choose top (or bottom) 10% of your plants with this trait and allow them to reproduce by transferring pollen between them
- Continue for MULTIPLE generations
- Should observe **directional selection**
  - Results in more plants at one end of the phenotypic range
- Controls
  - Type of plant, age of plant when trait was measured, same method used to transfer pollen

Investigation 2: Hardy-Weinberg

- Hardy-Weinberg equilibrium applies to a population that is NOT evolving
  - Frequency of alleles in the population remains the same generation after generation IF the following 5 conditions are met
    - No mutations
    - No gene flow
    - No genetic drift (large population size)
    - No natural selection (no advantages/disadvantages to trait)
    - Random mating

- **Directional Selection**
  - Ex: Individuals homozygous for sickle cell do not survive to reproduce
  - Shifts allele frequency
    - More "A"; less "a"
      - Frequency of p increased while q decreased
    - But "a" NOT eliminated because still carried in heterozygotes
- **Heterozygote Advantage**
  - Ex: Carriers of sickle cell allele are more resistant to malaria
  - Shifts allele frequency
    - "A" more common than at first but not as common as in directional selection; "a" less common than at first but more common than in directional selection
  - Also known as **stabilizing selection**
- **Genetic Drift**
  - Chance events cause unpredictable fluctuations in allele frequencies
  - Effects are most pronounced in SMALL populations
- Calculations
  - Be able to calculate frequency of each allele AND genotype
    - Start by labeling p, q, p^2, q^2, 2pq
    - Start with q^2 (frequency of recessive genotype/phenotype)

Investigation 3: BLAST

- Compare commonalities in gene and/or amino acid sequence among different organisms to determine evolutionary relationship
- Species that show more similarity will be placed closer together on cladogram
- Each branching point on the cladogram represents a species that lived in the past and gave rise to two or more species that came later
  - In other words, it's a **common ancestor** to the species that come later

Investigation 4: Diffusion and Osmosis

- Water moves from high to low water potential
  - Higher solute concentration = lower water potential
- Cells with highest surface area: volume ratio will have faster rate of diffusion
  - To calculate ratio for a block:
    - Surface area = L x W x 6 (which is number of sides)
    - Volume = L x W x H
    - Ratio = Surface area / volume

- Calculations
  - Be able to calculate solute potential
    - Ψ_S = iCRT
      - i = ionization constant (1 for sucrose, 2 for NaCl)
      - C = molarity
      - R = pressure constant (given as 0.0831)
      - T = temp (convert C to K by adding 273)
  - Be able to calculate water potential
    - $\Psi = \Psi_P + \Psi_S$
      - Ψ = water potential; Ψ_S = solute potential ; Ψ_P = pressure potential
      - In an open container, water potential = solute potential
  - Use that to determine which direction water will move
    - The bigger the difference in water potential between cell and solution, the bigger the movement of water
  - Be able to explain procedure and complete calculations used to determine potato's molarity
    - Given mass of potato before and after being placed in various concentrations of solution, calculate percent change in mass
      - % change = [(Final Mass – Initial Mass) / Initial Mass] x 100%
    - Graph % change (dependent variable on y axis) vs molarity/concentration of solution (independent variable on x axis) and draw line of best fit
    - To determine molarity of potato, find where line of best fit crosses x-axis (**isotonic**)
      - Potato will lose mass in a **hypertonic** environment (- % change in mass)
      - Potato will gain mass in a **hypotonic** environment (+ % change in mass)
        - The greater the weight gain, the lower the molarity of solution

Investigation 5: Photosynthesis

- Measure rate of photosynthesis based on amount of oxygen produced, causing disks to float
  - First have to sink the disks by removing all gases (vacuum)
  - Need source of carbon dioxide (sodium bicarbonate solution)
  - Need light
    - Absorb red and blue light (Fastest rate of photosynthesis)
    - REFLECT green light (Slowest rate of photosynthesis)

- Possible independent variables
  - Wavelength of light (various colors)…this is what we tested
  - Light intensity, temperature, carbon dioxide levels, type of leaf, distance from light
- Dependent variable = 1 / ET_{50} (time required for 50% of leaf disks to float)
  - Found by graphing # of floating disks vs time and then finding median
- Controls
  - Water control group (no bicarbonate solution showing no disks should be floating if no photosynthesis happening)
  - Type of leaf, distance from light, wattage of light, temperature, etc.
- Predictions
  - What would happen to the floating disks after the light was turned off and why?
    - Disks would start sinking
    - Using oxygen during respiration (no photosynthesis happening)

Investigation 6: Cellular Respiration

- 3 ways to measure respiration
  - Oxygen consumption (what we measured)
  - Carbon dioxide production
  - Amount of glucose consumed or energy released during respiration
- Used respirometer to measure change in volume of oxygen
  - Submerged in water (also helped keep temp constant)
- Need to limit following variables
  - CO_2 released during respiration so the only gas we are measuring is O_2
    - Use KOH to absorb CO_2 and form solid precipitate
  - Temperature and pressure changes since PV = nRT
    - Use control group of beads with same volume to account for any changes due to temp and pressure rather than respiration
- To determine rate of oxygen consumption
  - Graph oxygen consumed (y-axis) vs time (x-axis)
  - Find slope ($\Delta Y / \Delta X$)
- Results
  - Germinating seeds consume more oxygen than nongerminating seeds
  - Seeds at higher temperature consume more oxygen than those at lower temperature

Investigation 7: Cell Division: Mitosis and Meiosis

- Part A: Mitosis
  - Goal: Determine if lectin affects number of cells undergoing mitosis in onion root cells
  - Count number of cells in mitosis vs interphase
  - Compare counts in treated cells vs control
  - Perform Chi-Square analysis to determine significance of any differences
    - Null hypothesis: There is NO statistical difference between the number of cell in mitosis in the control vs treated group
      - Any differences are due to chance
    - Contingency box has 2 columns
      - Interphase and Mitosis
    - To calculate expected values
      - Find PERCENTAGE of cells in interphase vs mitosis in CONTROL group
      - MULTIPLY percentages by total number of cells in TREATED group
    - ACCEPT null if calculated chi square value is LESS THAN critical
    - REJECT null if calculated chi square value is GREATER THAN critical
      - There is a significant difference between number of cells in mitosis in control vs treated group
      - Evidence that treatment influences mitosis

- Part B: Meiosis
  - Goal: Determine frequency of crossover-produced ascospores to determine relative distance separating the gene locus and the centromere
  - Count asci genotype combinations
    - 4 black : 4 tan or vice versa indicates NO crossover
    - 2b:2t:2b:2t, 2b:4t:2b, or vice versa indicates CROSSOVER (recombinant asci)
  - Recombinant frequency = (recombinant asci / total asci) x 100
  - Divide recombinant frequency by 2 to determine map units

Investigation 8: Biotechnology: Bacterial Transformation (pGLO lab)

- **Transformation** = transfer of genetic info into cell by direct uptake of DNA from environment
- Use *E. coli* bacteria because
  - Unicellular; no nuclear envelope; plasmids; reproduce quickly; easily grown

- To transform bacteria
    - Keep competent *E. coli* cells (means they can accept DNA from another source) on ice
        - Holes in membrane, in calcium chloride solution
    - Add pGLO plasmid DNA
    - Incubate on ice and then heat shock by quickly transferring to heat block and back to ice
        - Opens pores in cell membrane so DNA can enter
    - Add nutrient broth (bacteria's food)
- Need control with no plasmid added
- To determine if transformation was successful
    - 3 +DNA and 3 -DNA plates
        - Control plates with just LB
            - Show bacteria are viable
        - LB amp plates
            - Contain ampicillin that will kill bacteria that do NOT contain plasmid (and were therefore not transformed)
        - LB amp ara plates
            - Transformed bacteria will glow under UV light
            - Arabinose is necessary to turn gene ON
                - Genes can be turned on and off so energy is not wasted making unnecessary proteins/enzymes

Investigation 9: Biotechnology: Restriction Enzyme Analysis of DNA

- Restriction Enzymes
    - Cut DNA at specific restriction sites (palindrome)
        - May create "sticky ends"
        - Ligase can be used to join DNA from different sources cut with same restriction enzyme
            - Genetically engineer plasmids to contain certain genes
- DNA Mapping
    - DNA cut at specific sequences with restriction enzymes can be separated using gel electrophoresis
    - Size of fragments (RFLPs) can then be compared
- Gel Electrophoresis
    - Separates fragments based on size
        - Smaller fragments move faster and therefore farther in given time period
    - Need electricity and buffer (and tracking dye, and ladder of known sizes)
        - DNA is negative so it is loaded into wells on negative site
        - Attracted to positive end when electricity is added

Investigation 10: Energy Dynamics

- Create a model ecosystem to estimate net primary productivity of plants and flow of energy to larvae
    - Estimate primary productivity by determining mass of plants "wet" and then "dry" (dehydrated)
    - Estimate energy flow between producers and consumers
        - Find mean mass of plant before and after consumer feeds on it
        - Find mean mass of larvae before and after consuming plant
        - Find mass of frass (waste)
- Focus is biomass (rather than rate of gas consumption/production as in photosynthesis/respiration labs)
- Production efficiency = net secondary production / assimilated energy (does not include waste)

Investigation 11: Transpiration

- Main driving force for movement of water from root to shoot is transpiration
    - When water evaporates from plant, it "tugs" water up xylem
        - Water sticks to water by cohesion and to plant cell walls by adhesion
- **Rate of transpiration INCREASES with**
    - **Increased air movement (more wind)**
    - **Decreased humidity (less moisture)**
    - **Increased light intensity**
    - **Increased temperature**
- Method 1
    - Measure entire plant's mass every day for about a week
        - Must seal bag around root so only water loss is through leaves
        - Remove any flowers or buds
        - If any leaves fall off, include that in mass each time
    - Need one plant as control and another affected by ONE variable
        - Change in temp, humidity (mist it), light intensity, etc.
    - To compare treatments
        - Could calculate percent change in mass
        - To determine total surface area of leaves
            - Find surface area of ONE leaf by tracing it on grid paper and then find its mass
            - Determine conversion ratio (SA/g)
            - Find mass of all leaves and apply conversion ratio to determine approximate surface area for all leaves

- Method 2
    - Use potometer to track water loss of only part of a plant inserted into water-filled tube with pipette on other end
    - As water evaporates from leaves, water is pulled down pipette
- Results
    - Increased water loss will decrease plant's weight
    - If transpiration is slowed, less water loss, less weight change
    - Higher leaf surface area = greater rate of transpiration
    - More stomata = more water loss
    - Increase in environment's water potential slows evaporation; decrease in water potential increases evaporation

Investigation 12: Fruit Fly Behavior

- Create choice chamber to determine flies' response to chemicals, light, and gravity
    - Response toward variable is positive taxis; away is negative taxis
- Put control (water) on cotton ball at one end and chemical at other end
- Eliminate other variables
    - No extra light shining on either end
    - Tube horizontal to eliminate effects of gravity
- Count flies at each end and perform chi square analysis to determine significance

Investigation 13: Enzyme Activity

- Investigating rate at which hydrogen peroxide is converted to water and oxygen
    - Enzyme peroxidase is used to speed up reaction
- Guaiacol is used to determine how much oxygen is produced
    - More oxygen produced = darker brown color
        - Can be compared to color chart or measured with spectrophotometer
- Calculate rate of reaction by graphing absorbance vs time and then finding slope of constant linear portion of curve
- Next investigate factors that affect rate of reaction
    - pH, temperature, substrate concentration, enzyme concentration
        - Turnip peroxidase tends to work best at a pH of 5
        - Reaction rates typically increase with increase in temp, substrate and enzyme concentration TO A POINT